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ORIGINAL ARTICLE

Statistical approaches to evaluate the aquatic ecosystem qualities of a significant mining area: Emet stream basin (Turkey)

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Abstract Emet Stream is one of the most important branches of Uluabat Lake (Ramsar area) and also one of Turkey's most important river systems. In addition to the geologic structure of the basin, Harmancik Chromium Mines are one of the most important inorganic pollution sources for the basin and also for Uluabat Lake. In the present study, water, sediment and fish (*Squalius cii*, *Capoeta tinca* and *Barbus oligolepis*) samples were collected seasonally from eight stations (one of them was on the Kinik Stream where the Harmancik Chromium Mines is located on and one of them was on the Dursunbey Stream) on the Emet Stream Basin. Some limnological parameters (pH, conductivity and total hardness) in water and Cr, Ca, Mg, Ni and Mn levels in biotic (muscle, gill, liver and kidney tissues of fishes) and abiotic (water and sediment) components of basin were determined to evaluate the effects of Harmancik Chromium Mines on the system. Cluster Analysis, Factor Analysis, Pearson Correlation Index, One Way Anova Tests, Scatter dot Comparing and Matrix plot Distribution Diagrams were applied to the results in order to estimate the data properly. Water samples were evaluated according to the water quality criteria for Turkey and sediment samples were evaluated according to the sediment quality criteria. According to data obtained, statistically significance differences were identified between Kinik and Emet Streams according to Cr and Ni accumulations in water and sediment. Chromium levels of Kinik Stream were extremely higher an average of 153 times for water and 10 times for sediment than uncontaminated stations. Water and sediment quality of Emet Stream were decreasing after falling the Kinik Stream and increasing after falling the Dursunbey Stream. It was also determined that, the amount of chromium in muscle tissues of three fish species were much higher than the limit value of 0.15 mg kg^{-1} that FAO identified.

Keywords Emet stream · Chromium · Water quality · Sediment quality · Statistical techniques

Introduction

Chromium is a naturally occurring element found in rocks, animals, plants, soil, and in volcanic dust and gases. Chromium is present in the environment in several different forms. Chromium enters the air, water, and soil mostly in the chromium III and VI forms as a result of natural processes and human activities. Although most of the chromium in water binds to dirt and other materials and settles to the bottom, a small amount of chromium can dissolve in the water (ATSDR 2000). The most significant

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anthropogenic point sources of chromium in surface waters and ground waters are the wastewaters from electroplating operations, leather tanning industries and textile manufacturing. In addition, deposition of airborne chromium is also a significant nonpoint source of chromium in surface water (Fishbein 1981).

At first, crocoite from Russia was the main source, but in 1827, a larger chromite deposit was discovered near Baltimore, United States. This made the United States the largest producer of chromium products until 1848 when large deposits of chromite where found near in Harmancik, Turkey (National Research Council 1974). Therefore, Harmancik Chromium Mines have a special importance not only for Turkey but also for the entire world. There are nine mines with a total of 704,055 tons of reserves and an annual production capacity of 20,000 tons in the Harmancik district and the region has an important place in the world's production of chromium (Mining Specialization Commission Report 2001).

Uluabat Lake, which has an important biological diversity and located on the migration route for many bird species and a Ramsar area, is one of the most important wetlands not only for Turkey but also for Europe and the Middle East (Magnin and Yarar 1997). Emet Stream, which is one of the most important branches of Uluabat Lake and also one of Turkey's most important river systems, is under pressure of Harmancik Chromium Mines by means of Kinik Stream and carries all the pollution factors directly into the Uluabat Lake.

Capoeta tinca (Heckel 1843) that is an endemic species for Turkey, and *Barbus oligolepis* Battalgil 1941 are living in subtropical climate freshwaters as benthopelagic fishes (Berg 1964; Turan et al. 2006; Turan et al. 2009). *Squalius cii* (Richardson 1857) is only found on Lesbos and in the streams flowing into the northern Aegean in Turkey. The species is locally threatened by pollution, water abstraction and drought and only survives in a few narrow areas with limited populations due to heavy industrial pollution (IUCN 2011). So the determination of bioaccumulation levels in especially metabolically active tissues (liver and kidney) of *S. cii* has a critical importance for a well estimation about the situation of species in the future.

The aim of this study was to evaluate the effects of Harmancik Chromium Mines on the Emet Stream Basin by determining some limnological parameters (pH, conductivity and total hardness) in water and Cr, Ca, Mg, Ni and Mn (interactive with chromium) levels in biotic (muscle, gill, liver and kidney tissues of *S. cii, C. tinca* and *B. oligolepis*) and abiotic (water and sediment) components of Emet Stream Basin, and by using some statistical techniques.

Materials and methods

Study area and collection of samples

The study area and selected stations on the Emet Stream Basin were given in Fig. 1. Localities and geographic information of stations are given in Table 1. The S1 station is located on the source of Emet Stream and away from any domestic and industrial pollution. The S2 station is located on the Emet Stream and close to a large settlement area but away from any point of organic and inorganic pollution. The S3 station is located on the Emet Stream and exposed to a significant agricultural and domestic discharges. S4 station was located on the Emet Stream after the discharge of Boron Facility by means of Gelenbek Stream. S5 station was located on the Kinik Stream and close to the Harmancik Chromium Mines. S6 station was located on the Emet Stream after the discharge of Chromium Facility by means of Kinik Stream. S7 station was located on the downside of Dursunbey Stream, where the rural settlement is dominated and away from any point inorganic pollution. And S8 station was located on the downside of Emet Stream, where could reflect all domestic, agricultural, industrial discharges due to the anthropogenic effects and natural—geologic effects that the system is exposed to and could reflect the pollution transported to the Uluabat Lake.

All samples were collected seasonally between November 2010 and August 2011. Water and sediment samples were collected from all stations by using sediment dipper, ekman grab and suitable containers. Fish samples were caught from four stations (S2, S4, S7, S8 stations) by using modified Honda generator (EM1000F, Honda Motors, Japan). Fishes that have high mobility spread throughout the entire basin. So the most appropriate stations with the structure of bottom stream and flow rate were chosen for the method of electro shocker.

Some metric characteristics of fishes were given in Table 2 (enough *B. oligolepis* species could not caught for element analysis in autumn and spring seasons).

Physical and chemical parameters

Conductivity and pH parameters were determined in situ using Multi-Parameter Measurement Device (HQ40d Portable Multiparameter Meter, Hach, US) during the field studies.

Chemical analysis

Fish samples were dried for 24 h at 105 °C. Sediment samples were dried for 3 h at 105 °C. Samples were placed (0.25 g of each sample) in Pyrex reactors of a CEM Mars Xpress 5 microwave digestion unit. $HClO_4$:HNO_3 acids of

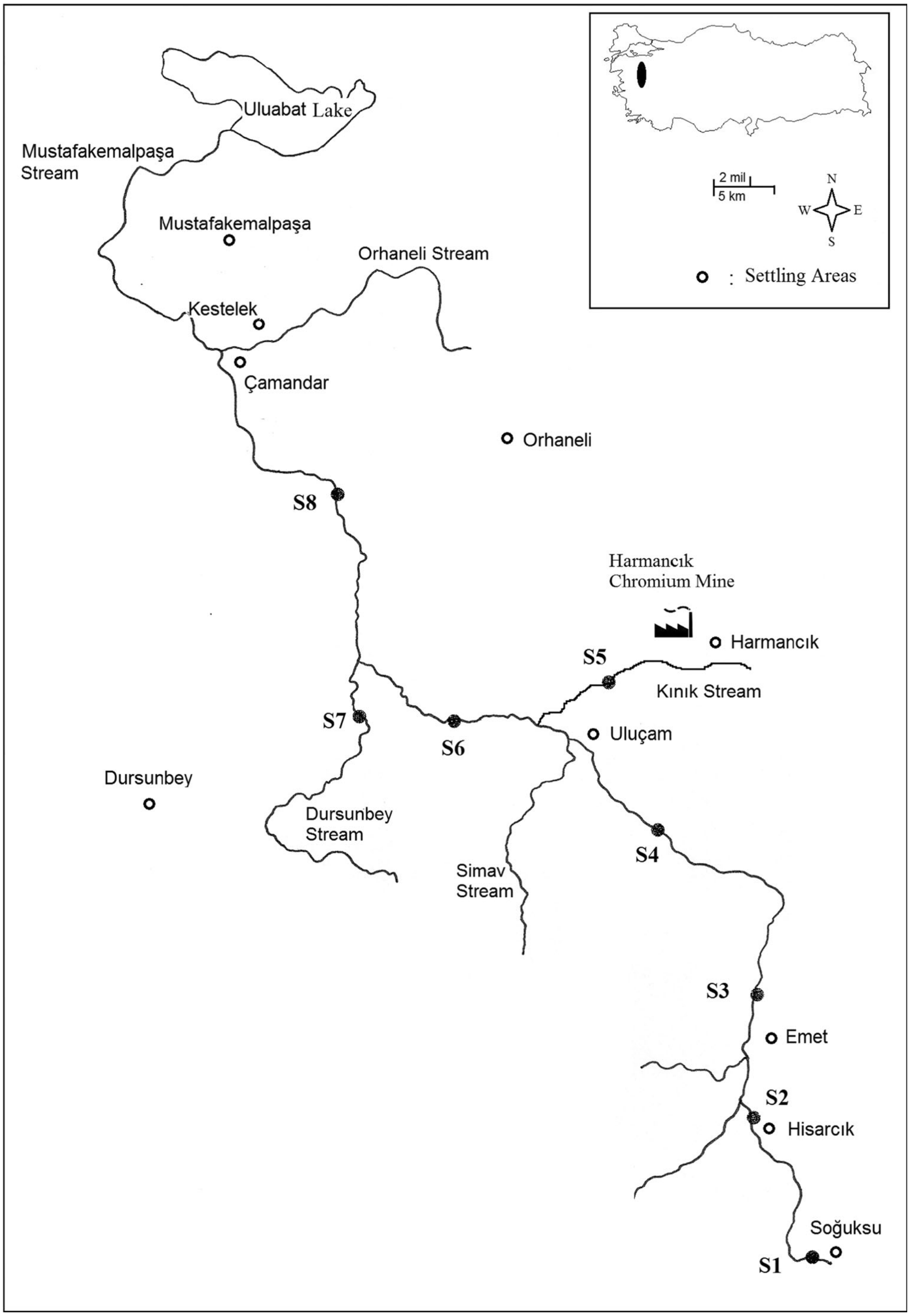

Fig. 1 Study area

1:3 proportions were inserted in the reactors respectively. Samples were mineralized at 200 °C for 30 min. Afterwards, the samples were filtered in such a way as to make their volumes to 100 ml with ultra-pure distilled water.

Water samples of 1 l that were taken at each sampling point were adjusted to pH two by adding 2 ml of HNO3 into each for determination of elements (Ni, Mn, Ca, Mg). For determination of total chromium in water, 100 ml from samples were transferred to a 250 ml beaker and 2 ml (1 + 1) of nitric acid and 1 ml (1 + 1) of hydrochloric acid were added. And then put on hot plate for evaporation to nearly dryness, making certain that the samples do not boil at 85 °C. Sample volume was come down to ~20 ml. Afterwards, the samples were filtered (cellulose nitrate, 0.45 μm) in such a way as to make their volumes to 50 ml with ultra-pure water.

Element levels were determined by Inductively Coupled Plasma-Optic Emission Spectrophotometric (Varian 720

Table 1 Coordinates and localities of each sampling sections of Emet stream basin

Stations	Locality	Coordinates		Altitude (m)
		Latitude	Longitude	
S1	Soğuksu village	39 09 835 N	029 17 396 E	838
S2	Aşağiyoncaağaç village	39 16 246 N	029 13 443 E	726
S3	Yağcik village	39 21 064 N	029 13 712 E	697
S4	Dereli village	39 28 111 N	029 15 179 E	555
S5	Kinik stream	39 39 895 N	029 02 263 E	509
S6	Ören village	39 37 091 N	028 55 323 E	357
S7	Dursunbey stream	39 37 454 N	028 43 445 E	305
S8	Düğüncüler village	39 40 321 N	028 43 450 E	270

Table 2 Metric characteristics of fishes

Fishes	Weight (gr)	Length (mm)		
		Standard	Fork	Total
S. cii ($n = 162$)				
Minimum	6	19	14	81
Maximum	247	237	259	271
Mean	50.5	132.2	147.3	159.8
SD	35.7	30.5	33.4	33.8
C. tinca ($n = 170$)				
Minimum	9	75	84	94
Maximum	250	233	257	281
Mean	59.9	141.7	156.7	172.6
SD	42.8	31.7	34.6	37.6
B. oligolepis ($n = 46$)				
Minimum	7	74	84	92
Maximum	107	187	206	228
Mean	24.8	108.8	119.7	132.3
SD	21.3	26.9	28.7	31.8

n number of samples, *SD* standard deviation

ES) method. The element analyses were recorded as means triplicate measurements (ASTM 1985; EPA METHOD 2007; EPA METHOD 2001).

Total water hardness

Total permanent water hardness was calculated by the following formula:

"Total Permanent Hardness = Calcium Hardness + Magnesium Hardness"

The calcium and magnesium hardness is the concentration of calcium and magnesium ions expressed as equivalent of calcium carbonate. The molar mass of $CaCO_3$, Ca^{2+} and Mg^{2+} are respectively 100.1 g/mol, 40.1 g/mol and 24.3 g/mol.

The ratio of the molar masses is:

$$\frac{M_{CaCO_3}}{M_{Ca}} = \frac{100,1}{40,1} = 2,5, \qquad \frac{M_{CaCO_3}}{M_{Mg}} = \frac{100,1}{24,3} = 4,1$$

So total permanent water hardness expressed as equivalent of $CaCO_3$ were calculated by the following formula (Younger 2009):

$$[CaCO_3] = 2,5[Ca^{2+}] + 4,1[Mg^{2+}].$$

Statistical analysis

Cluster Analysis and Matrix plot Diagrams was applied to the results by using the Past package program. One Way Anova Test, Pearson Correlation Index, Factor Analysis and Scatter dot Diagrams were applied to the results by using the SPSS 17 package program.

Results and discussion

Seasonal pH, conductivity and total hardness levels of Emet Stream Basin were given in Table 3. Major deviations from the optimum pH levels were not detected. In general, water of Emet Stream Basin had alkaline character in spring and summer seasons and also had very hard water character. According to Water Pollution Control Regulations in Turkey, all stations except S5 station (III. Class in winter season) had I. Class water quality in terms of pH values (SKKY 2004). The highest conductivity levels were measured in S5 station in all seasons except summer season.

Annual averages of element levels determined in water and sediment of Emet Stream Basin and the results of One Way Anova Test that compares the element accumulations of stations were given in Table 4. According to the results of One Way Anova Test, statistically significance differences were identified for water between S5 station with all stations in terms of Mg (except S6), Ni, Mn and Cr levels; between S8 station with S1 and S2 stations in terms of Mn levels; and between S6 station with S7 station in terms of Cr levels ($p < 0.05$). Also statistically significance differences were identified for sediment between S5 station with all stations in terms of Mg, Ni and Cr levels; between S4 station with all stations in terms of Ni levels; between S6 station with all stations (except S8) in terms of Ni and Cr levels; between S7 station with all stations in terms of Cr levels; between S5 station with S1, S2 and S8 stations in terms of Mn levels; and between S4 station with S1 and S2 stations in terms of Mn levels ($p < 0.05$).

According to seasonal variations of element levels, significant increases of Ca accumulations in water and

Table 3 Seasonal pH, conductivity and hardness values detected in water of Emet stream basin

St.	Seasons															
	Autumn				Winter				Spring				Summer			
	pH	Cond. (µS/cm)	Hardness		pH	Cond. (µS/cm)	Hardness		pH	Cond. (µS/cm)	Hardness		pH	Cond. (µS/cm)	Hardness	
			$CaCO_3$ (mg l^{-1})	Indi.			$CaCO_3$ (mg l^{-1})	Indi.			$CaCO_3$ (mg l^{-1})	Indi.			$CaCO_3$ (mg l^{-1})	Indi.
S1	7.4	677	141	*2	7.5	284	243	*3	7.6	673	360	*3	7.9	833	549	*3
S2	7.8	761	170	*2	7.3	547	437	*3	7.9	675	403	*3	8	973	621	*3
S3	7.5	680	247	*3	7.4	463	351	*3	8.0	636	389	*3	8	1070	644	*3
S4	7.3	813	338	*3	7.5	467	344	*3	8.3	680	386	*3	8.2	1172	677	*3
S5	7.5	712	297	*3	8.6	610	345	*3	8.0	688	518	*3	8.5	1021	531	*3
S6	7.4	682	319	*3	7.3	468	371	*3	8.7	616	372	*3	8.3	928	518	*3
S7	8.4	413	105	*1	7.3	156.3	117	*1	8.5	273	144	*2	8.5	561	336	*3
S8	8.3	625	232	*3	7.2	355	294	*3	8.6	524	359	*3	8.6	833	487	*3

St. stations, *Cond.* conductivity, *Indi.* indications; *1: moderately hard water, *2: hard water, *3: very hard water

sediment and significant increases of Mg accumulations in water were determined in summer season and no significant seasonal changes were determined for Ni, Mn and Cr accumulations in both water and sediment.

Chromium occurs naturally in the Earth's crust and continental dust is the main source of exposure to natural chromium present in the environment. Rain helps to remove chromium from air and washes chromium compounds out of many soils so that it eventually moves into the surface and groundwater (Fishbein 1981; ATSDR 2000). Determination of no significant increases in levels of chromium, magnesium, nickel and manganese in rainy seasons such as spring and autumn reflect that, environmental interactions could not effect Cr levels and its compounds as much as discharges from a point source like Harmancik Chromium Mines.

The scatter dot graphics which compare the Cr, Mn and Ni levels of water and sediment with water and sediment quality criteria are given in Fig. 2.

According to the Water Pollution Control Regulation in Turkey (SKKY 2004), S1, S2, S3, S4 and S7 stations had I. Class (<0.02 mg l^{-1}) water quality, S6 and S8 stations had II. Class (0.02–0.05 mg l^{-1}) water quality and S5 station had IV. Class (>0.2 mg l^{-1}) water quality in terms of Cr levels. S1, S2, S3, S4, S6, S7 and S8 (except spring season) stations had I. Class (<0.1 mg l^{-1}) water quality and S5 station had II. Class (0.1–0.5 mg l^{-1}) water quality in terms of Mn levels. S1, S2, S3, S4 and S7 stations had I. Class (<0.02 mg l^{-1}) water quality, S6 and S8 stations had II. Class (0.02–0.05 mg l^{-1}) water quality and S5 station had IV. Class (>0.2 mg l^{-1}) water quality in terms of Ni levels. Chromium levels in natural waters are quite low and have to be about 0.001–0.002 mg l^{-1} in uncontaminated waters (Moore and Ramamoorthy 1984). The Cr levels determined in water were over these limit values in all stations. These results could indicate that, in addition to the pressure of Harmancik Chromium Mines, geologic structure of Emet Stream Basin is also an important factor on chromium accumulations in water.

According to the sediment quality criteria specified by (MacDonald et al. 2000), although the Cr levels of sediment had exceeded the threshold effect level (TEL, 37.3 mg kg^{-1}) in S1, S2, S3 and S4 stations and had exceeded the lowest effect level (LEL, 26 mg kg^{-1}) in S7 station, chromium accumulations have not reached the critical levels in these stations yet. But Cr levels in sediment of S5, S6 and S8 stations have reached critical levels and outrun the minimal effect threshold value (MET, 55 mg kg^{-1}). Ni levels of sediment have not exceeded the lowest effect level (LEL, 16 mg kg^{-1}) in S1 and S2 (except autumn season) stations, has not exceeded the threshold effect level (TEL, 18 mg kg^{-1}) in S7 (except autumn season) station, has not exceeded the minimal effect threshold value (MET, 35 mg kg^{-1}) in S3 (except autumn season) station and has slightly exceeded the minimal effect threshold value in S4 station. But Ni levels in sediment of S5, S6 and S8 stations have exceeded even the minimal effect threshold value (MET, 35 mg kg^{-1}). Accumulations of chromium and nickel in sediment of Emet Stream Basin were considerably higher than the accumulations in water especially in contaminated stations (S5, S6 and S8).

In a study performed in an industrial development area in India, minimum and maximum chromium and nickel levels determined in sediments of a few lakes were 62–172 mg/kg and 24–234 mg/kg respectively (Govil et al. 2012). If we compare the present results with this study, although maximum Ni level in sediment of Indian lakes was higher than Emet Stream Basin, chromium levels detected in sediment of S5 station were an average of five

Table 4 Seasonal averages of element levels determined in water and sediment

Stations	Water (mg l^{-1})					Sediment (mg kg^{-1})				
	Ca	Mg	Ni	Mn	Cr	Ca	Mg	Ni	Mn	Cr
S1										
(Min)	17.23333	13.61667	0.009833	0.007001	0.002333	48986.67	5758.407	12.86667	178.4313	36.06667
(Max)	144.856	46.97228	0.011827	0.0115	0.002499	70445.27	18006.67	16.40533	265.6	42.2
(Mean)	**84.23289^{a}**	**28.24806^{a}**	**0.010811^{a}**	**0.009842^{a}**	**0.002412^{a}**	**60235.97^{a}**	**10331.2^{a}**	**15.23185^{a}**	**217.8894^{a}**	**39.74232^{a}**
(SD)	53.05757	13.93729	0.000848	0.002144	0.0001	8782.871	5686.306	1.60156	43.26691	2.626466
S2										
(Min)	17.51667	28.63333	0.010417	0.012448	0.003383	41666.67	4829.347	11.78	203.8853	39.97267
(Max)	168.2968	49.64585	0.013544	0.023872	0.003605	53333.33	14918.73	31.66667	253	43.8152
(Mean)	**106.9751^{a}**	**35.0954^{a}**	**0.011657^{a}**	**0.017955^{a}**	**0.003495^{a}**	**45866.59^{a}**	**8829.803^{a}**	**17.73381^{a}**	**221.7525^{a}**	**41.8728^{a}**
(SD)	64.03651	9.767632	0.001358	0.005511	0.00011	5276.429	4391.421	9.363718	21.60857	1.763023
S3										
(Min)	43.61667	24.16667	0.009833	0.019833	0.003365	39186.67	5288.9	23.35333	261.7432	38.35533
(Max)	174.0785	51.1415	0.014086	0.071264	0.003561	59728.92	11378.04	39.33333	394.4586	41.39
(Mean)	**106.9074^{a}**	**34.7916^{a}**	**0.012163^{a}**	**0.040851^{ac}**	**0.003457^{a}**	**46458.15^{a}**	**7530.652^{a}**	**29.94279^{a}**	**317.7845^{abc}**	**39.59323^{a}**
(SD)	53.40378	11.72888	0.002122	0.021974	0.0001	9261.628	2655.524	7.722602	55.86443	1.280805
S4										
(Min)	76.15	23.7	0.010667	0.019167	0.004079	42172.47	9634.713	51.83573	314.0464	42.65125
(Max)	174.852	59.45988	0.0135	0.040212	0.004673	61805.13	13947.58	61.93333	429.5847	43.86667
(Mean)	**114.3361^{a}**	**37.31296^{a}**	**0.012176^{a}**	**0.030451^{ac}**	**0.004399^{a}**	**48771.07^{a}**	**11787.41^{a}**	**57.21865^{b}**	**360.3019^{bc}**	**43.47891^{a}**
(SD)	42.35196	15.59768	0.001437	0.009269	0.000254	8834.809	1853.791	5.197106	55.15236	0.558693
S5										
(Min)	17.48333	61.81667	0.576467	0.328333	0.42685	21166.67	97619.4	171.4247	385.2133	311.0031
(Max)	42.61477	108.863	0.779333	0.43245	0.500886	29972.8	127203.1	201.0646	517.4741	355.7085
(Mean)	**32.20147^{a}**	**84.33014^{b}**	**0.722475^{b}**	**0.388979^{b}**	**0.462054^{b}**	**24150.18^{b}**	**108872.3^{b}**	**187.6372^{c}**	**437.9562^{c}**	**332.1405^{b}**
(SD)	11.12414	25.66677	0.098025	0.044971	0.030375	3959.623	12883.71	13.89171	56.21541	18.3098
S6										
(Min)	52.5	41.58333	0.032261	0.025556	0.036489	18091.67	17701.73	99.66667	249.2	120.8138
(Max)	89.75548	72.62915	0.042667	0.044	0.041225	35340	25209.71	117.8031	439.0498	133.3553
(Mean)	**73.54199^{a}**	**52.40355^{ab}**	**0.037378^{a}**	**0.038598^{ac}**	**0.038641^{c}**	**23655.59^{b}**	**20295.71^{a}**	**107.8726^{d}**	**318.204^{abc}**	**125.5371^{c}**
(SD)	16.19437	13.81451	0.00459	0.008786	0.001982	8108.543	3387.588	8.28746	83.33334	5.454916
S7										
(Min)	14.9	9.441667	0.008833	0.014167	0.001058	22603.01	16356.35	14.90483	349.6667	20.21913
(Max)	67.26897	41.06707	0.013284	0.025074	0.001147	25580	18173.49	28.26667	411.5819	23.51323
(Mean)	**38.2834^{a}**	**20.33712^{a}**	**0.010663^{a}**	**0.019419^{ac}**	**0.00111^{a}**	**24346.02^{b}**	**16918.13^{a}**	**19.70081^{a}**	**366.2063^{bc}**	**22.29704^{d}**
(SD)	21.80543	14.25884	0.001967	0.004481	0.0001	1479.461	852.0297	6.12696	30.27895	1.451976
S8										
(Min)	43.23333	31.18333	0.021564	0.008326	0.020555	16480	14106.67	82.89333	216.193	107.1403
(Max)	80.1488	70.7654	0.049667	0.149776	0.023406	29869.95	21773.14	101.5106	330.647	117.4744
(Mean)	**65.61003^{a}**	**44.49493^{a}**	**0.034262^{a}**	**0.083109^{c}**	**0.021926^{ac}**	**21607.14^{b}**	**18391.77^{a}**	**89.96825^{d}**	**291.359^{ab}**	**111.9739^{c}**
(SD)	16.12157	18.43656	0.013821	0.058179	0.001183	6294.303	3487.639	8.547165	53.71086	4.243406

The values marked with different letters in the same column are statistically different ($p < 0.05$)

min minimum, *max* maximum, *mean* seasonal average, *SD* standard deviation

times higher than Indian lakes in industrial area. In another study performed in sediment of Yangtze River Basin estuary in China, average chromium level was recorded as 52.1 mg/kg (An et al. 2009). This stated value in one of the most contaminated area of the world was extremely lower than the accumulations detected in the Kinik Stream.

Annual and stational averages of element bioaccumulations in fish tissues and the results of One Way Anova

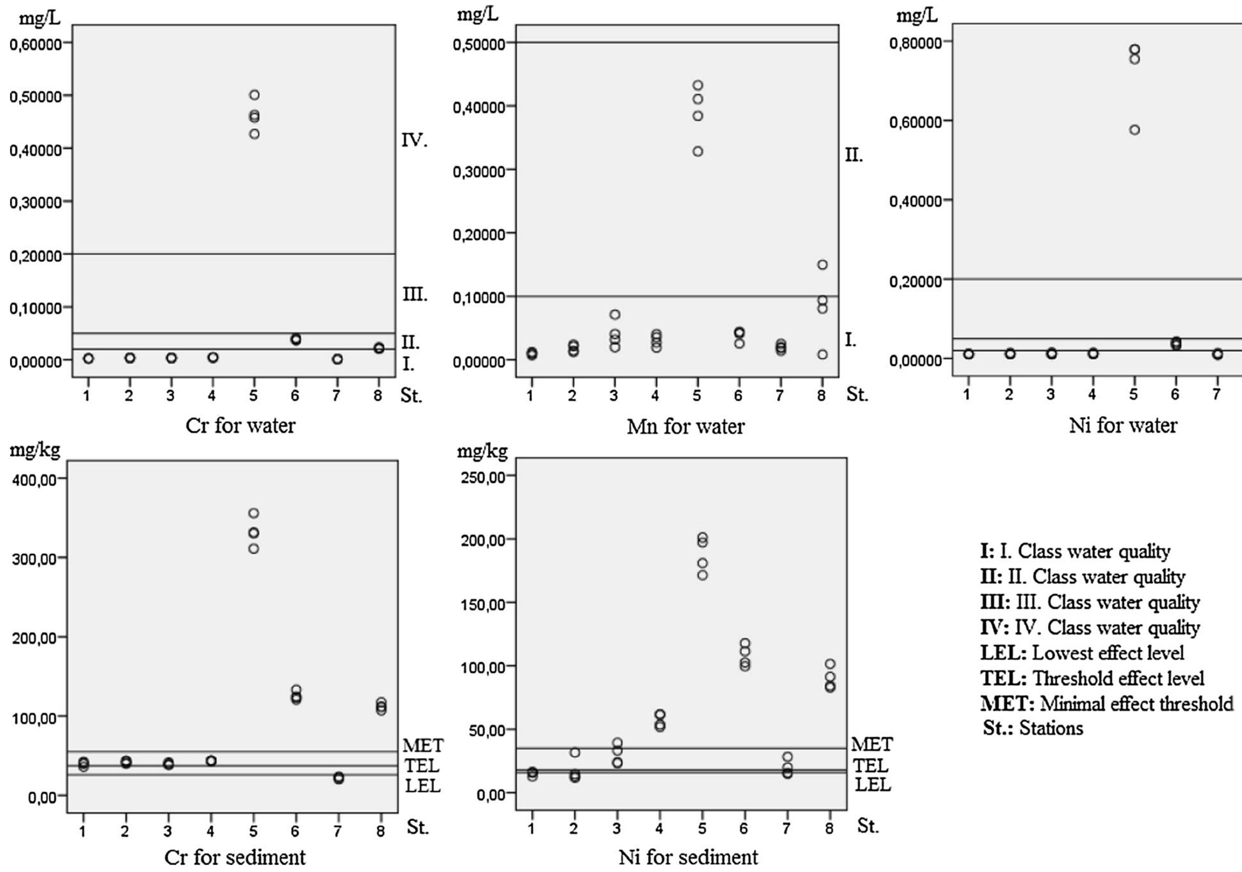

Fig. 2 *Scatter dot* Comparing Diagrams for water and sediment

Test that compares the element bioaccumulations of tissues were given in Table 5. Annual averages of chromium and nickel levels in tissues of three fish species indicating the stational differences and presented by using Matrix plot Distribution Diagrams were given in Fig. 3. Cr and Ni bioaccumulations of fish tissues detected in S2 and S7 stations (uncontaminated stations) were significantly lower than detected in S8 station (downside of Emet Stream and after from Kinik Stream discharge).

According to the results of One Way Anova Test, statistically significance differences were identified between gill tissues with all other tissues in terms of Ca and Mn bioaccumulations; and between muscle tissues with liver and kidney tissues in terms of Ni and Cr bioaccumulations in all three fish species ($p < 0.05$).

The metals (non-lethal concentrations) are accumulating in metabolically active tissues especially when the fishes exposed to heavy metals for a long time. The detoxification mechanisms of fishes are the same and the metals are primarily linked to the metallothionein protein which forms complexes with heavy metal ions and liver tissue that is detoxification organ of the fishes contains many of these proteins, so liver tissue accumulates the heavy metals significantly higher than other tissues. Kidney is also the secondary gateway for heavy metal detoxification in body and reduces the effects of metals (Kargin and Erdem 1992; Kalay and Erdem 1995; Ünlü et al. 1996; Vinodhini and Narayanan 2008; Lu et al. 2011). But as reported in many studies, the types of fishes, different physiological structures and exposures durations of heavy metals may affect to accumulate metals in different organs in different levels (Canli et al. 1998; Cid et al. 2001; Karadede et al. 2004; Mendil and Uluözlü 2007; Al-Weher 2008; Sen et al. 2011). According to the results of the present study, the highest bioaccumulations of Ni and Cr were determined in liver and kidney tissues because of high toxicity of these metals. The higher accumulation in liver and kidney may alter the levels of various biochemical parameters in these tissues. This may also cause severe liver and kidney damage and a primary impact on the health of fish (Mayers and Hendricks 1984; Ferguson 1989).

Gills are clearly in contact with water and gas exchange organ of the fishes. During this exchange, gill filaments are in contact with heavy metals. So heavy metal accumulations in water and gills show a close relationship and gill tissues are commonly used in toxicological studies

Table 5 Levels of metals in tissues of fishes

Stations	Tissues	Elements (mg kg^{-1})				
		Ca	Mg	Ni	Mn	Cr
S. cii	Muscle					
	(Mean)	**2571.86**^{a}	**827.130**^{a}	**0.972**^{a}	**0.393**^{a}	**0.378**^{a}
	(SD)	684.351	66.068	0.615	0.119	0.276
	Gill					
	(Mean)	**40701.97**^{b}	**1951.929**^{b}	**1.794**^{b}	**1.409**^{b}	**0.873**^{b}
	(SD)	7559.976	154.547	0.699	0.201	0.656
	Liver					
	(Mean)	**2931.15**^{a}	**1316.132**^{c}	**2.679**^{c}	**0.676**^{a}	**1.658**^{c}
	(SD)	1153.999	334.011	1.367	0.186	0.913
	Kidney					
	(Mean)	**3669.22**^{a}	**695.602**^{a}	**2.784**^{c}	**0.647**^{a}	**1.429**^{c}
	(SD)	710.012	78.888	1.391	0.181	0.954
C. tinca	Muscle					
	(Mean)	**10070.04**^{a}	**1064.32**^{a}	**1.591**^{a}	**1.037**^{a}	**0.960**^{a}
	(SD)	15472.544	474.224	0.602	0.631	0.486
	Gill					
	(Mean)	**39934.93**^{b}	**1976.64**^{b}	**2.270**^{ab}	**2.580**^{b}	**1.165**^{ab}
	(SD)	9679.846	420.038	0.439	0.509	0.429
	Liver					
	(Mean)	**2827.188**^{a}	**1454.86**^{ab}	**3.080**^{b}	**1.466**^{c}	**1.854**^{b}
	(SD)	1339.455	310.464	0.961	0.252	0.558
	Kidney					
	(Mean)	**5784.589**^{a}	**703.63**^{a}	**3.083**^{b}	**1.148**^{ac}	**1.696**^{b}
	(SD)	2577.328	221.489	1.083	0.331	0.601
B. oligolepis	Muscle					
	(Mean)	**7034.7**^{a}	**5271.9**^{a}	**1.042**^{a}	**0.327**^{a}	**0.374**^{a}
	(SD)	6121.591	7838.652	0.677	0.066	0.277
	Gill					
	(Mean)	**52654.1**^{b}	**4644.2**^{a}	**1.788**^{b}	**1.625**^{b}	**0.959**^{b}
	(SD)	17319.681	5116.06	0.773	0.298	0.619
	Liver					
	(Mean)	**6173.4**^{a}	**4631.9**^{a}	**2.711**^{c}	**0.962**^{c}	**1.619**^{c}
	(SD)	6753.303	6766.14	1.172	0.237	0.697
	Kidney					
	(Mean)	**13151.7**^{a}	**3827.7**^{a}	**2.636**^{c}	**0.670**^{ac}	**1.467**^{c}
	(SD)	14301.14	5926.6	1.444	0.080	0.709

The values marked with different letters in the same column are statistically different ($p < 0.05$)

(Amundsen et al. 1997; Canli and Atli 2003; Altindağ and Yiğit 2005). In the present study, highest bioaccumulations of Ca, Mg and Mn were determined in gill tissues and determination of lower bioaccumulations of Ca, Mg and Mn in metabolically active tissues reflects that, these elements are not toxic for fishes as much as Cr and Ni.

Muscle tissues of fishes are the only tissues that can be eaten and heavy metals accumulated in muscle tissues can easily pass to people. Therefore, some organizations such as FAO, WHO and EPA determined the limit values of heavy metals in muscle tissues of fishes. The amount of chromium in muscle tissues of three fish species were much higher than the limit value of 0.15 mg kg^{-1} that FAO (Food and Agriculture Organization) identified (FAO 1983). Muscle tissue does not contain an active structure of heavy metal binding, so using muscle tissues to estimate heavy metal levels in water may give incorrect results (Kayhan et al. 2009). In the present study, it was an expected result that, the lowest Cr and Ni bioaccumulations were determined in muscle tissues of all three fish species which have an inactive structure of heavy metal binding.

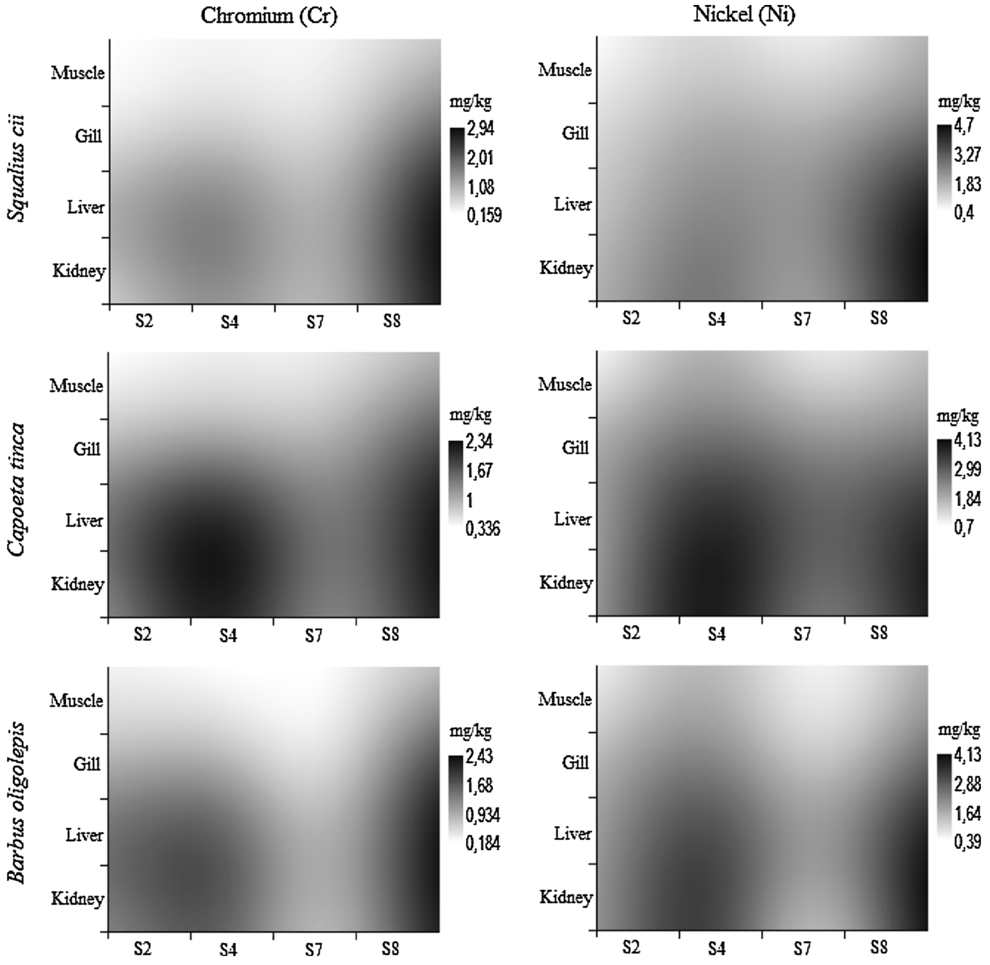

Fig. 3 Chromium and nickel bioaccumulations in tissues of fishes

Table 6 Pearson correlation index coefficients between metals and physicochemical parameters

Parameters	Water					Sediment					pH	Cond.	Hard.
	Ca	Ni	Mg	Cr	Mn	Ca	Ni	Mn	Mg	Cr			
Water													
Ca	1												
Ni	−389*	1											
Mg	007	673**	1										
Cr	−384*	982**	732**	1									
Mn	−351*	963**	712**	970**	1								
Sediment													
Ca	643**	−349	−176	−352*	−394*	1							
Ni	−396*	819**	771**	849**	836**	−564**	1						
Mn	−204	521**	571**	565**	558**	−319	570**	1					
Mg	−422*	982**	736**	979**	964**	−409*	848**	582**	1				
Cr	−380*	930**	794**	957**	942**	−471**	949**	527**	943**	1			
pH	−003	165	390*	212	236	−238	253	475**	231	240	1		
Cond.	466**	139	632**	189	149	392*	213	282	155	211	276	1	
Hard.	770**	131	643**	172	185	381*	188	210	146	215	243	762**	1

cond. conductivity, *hard.* hardness

$^{*}p < 0.05$, $^{**}p < 0.01$

Table 7 Extracted values of various FA parameters

Component	Extraction sums of squared loadings (un-rotated)			Rotation sums of squared loadings (rotated)		
	Total	% of Variance	Cumulative %	Total	% of Variance	Cumulative %
1	7.098	59.146	39.146	7.097	59.138	59.138
2	2.926	24.387	83.534	2.927	24.396	83.534

Levels of Cr and Ni in water and sediment of S7 (Dursunbey Stream) and S2 (upstream of Emet Stream) stations were determined significantly lower than the contaminated stations. As similar to accumulations in abiotic components of the basin, bioaccumulation levels of Cr and Ni in liver and kidney tissues of all three fish species caught from S7 to S2 stations were also lower than the fishes caught from S4 to S8 stations (after from discharges of chromium mines). These results show that, accumulations of toxic metals in biotic and abiotic components of aquatic ecosystems have a close relationship and Cr and Ni elements could be classified as limiting factor for the density of *S. cii*, *C. tinca* and *B. oligolepis* populations in the Emet Stream Basin.

The relationships between pH, conductivity, hardness and element levels in water and sediment were determined by the Pearson Correlation Index by using seasonal values ($n = 32$ for all parameters) and all determined significant relations were given in Table 6. It was found that, the relations between Ca levels hardness and conductivity; conductivity hardness; Cr levels Mg, Ni and Mn levels both in water and sediment were directly proportional ($p < 0.01$).

Calcium concentrations up to and exceeding 100 mg l^{-1} are common in natural waters, particularly groundwater. Magnesium is present in natural water usually at lower concentrations (from negligible to about 50 mg l^{-1} and rarely above 100 mg l^{-1}), so calcium based hardness usually predominates (National Research Council 1977). In this study, significant relations between Ca levels of water and hardness with a coefficient score of 0.770 ($p < 0.01$) were identified as similar to this information. Although, high Mg levels were determined after from especially S5 station, contents of Ca dominate and primarily effect on total hardness levels in water in the basin.

Chromite is a spinel mineral and can be represented by the “$X^{+2,\ +3}OY_2^{+3}O_3$” formula. The “X” cations in the formula can be Mg^{+2}, Mn^{+2} or Ni^{+3} and Cr^{+3} usually take the place of “Y” cations (Karsli 1994). So, high correlation coefficients between Cr with Mg, Ni and Mn were an expected condition.

Table 8 Parameter loadings of component matrix and rotated component matrix

Parameters	Component matrix		Rotated component matrix	
	1	2	1	2
Sediment chromium	0.979		0.979	
Sediment magnesium	0.971		0.970	
Water chromium	0.968		0.968	
Water manganese	0.957		0.956	
Water nickel	0.946		0.945	
Sediment nickel	0.929		0.929	
Water magnesium	0.819		0.826	
Sediment manganese	0.656		0.657	
Hardness		0.937		0.933
Conductivity		0.845		0.841
Water calcium		0.834		0.840
Sediment calcium		0.633		0.640

Recently, Factor Analysis (FA) is widely used for evaluating water and sediment quality of freshwater ecosystems (Najar and Khan 2012; Tokatli et al. 2012; Mao et al. 2013). In the present study, FA was used to determine the effective varifactors on Emet Stream Basin by using correlated variables. Uncorrelated variables were removed to increase the reliability of FA and a total of twelve variables were used to determine the varifactors. Result of KMO (Kaiser–Meyer–Olkin) measure of sampling adequacy test was 0.762 and this value means that, the sampling adequacy was in a good level (>0.7). Eigenvalues higher than 1 were taken as criteria to evaluate the principal components required to explain the sources of variance in the data.

The percentage variance was counted; cumulative percentage variance and component loadings (un-rotated and rotated) are given in Table 7. According to rotated cumulative percentage variance, two factors explained 83.53 % of the total variance.

The parameter loadings (>0.5) for two components before and after rotation are given in Table 8. (Liu et al. 2003) classified the factor loadings as “strong (>0.75)”, “moderate (0.75–0.50)” and “weak (0.50–0.30)”, according to loading values. Component plot in rotated space, which shows the related variables of two factors, is given in Fig. 4.

First factor (F1), named as “Chromium Mines Factor” explained 59.13 % of total variance and it was related to the variables of Cr, Mg, Mn and Ni values of water and sediment. All parameters except Mn in sediment were strong positively loaded with this factor, and also

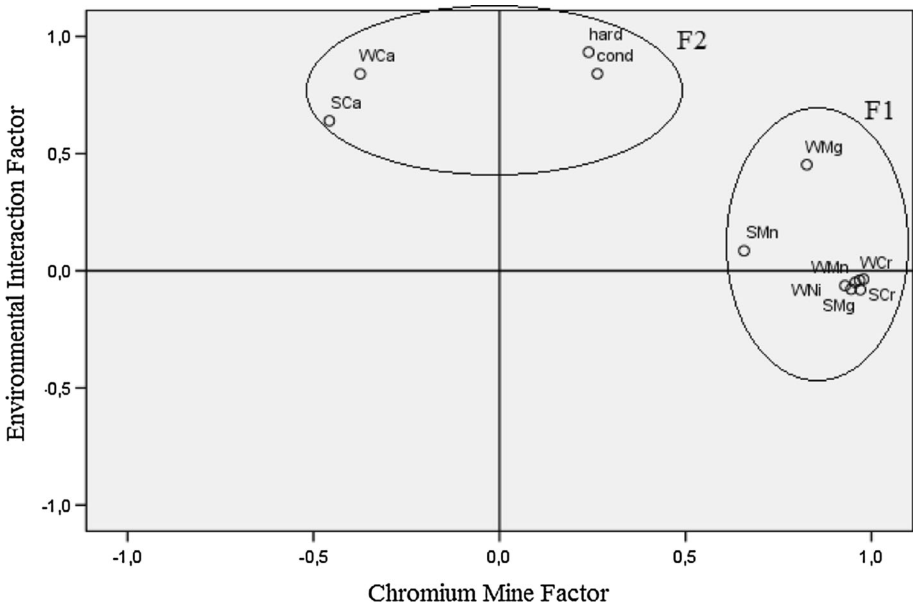

Fig. 4 Component *plot* in rotated space

parameter of Mn in sediment was moderate positively loaded with this factor.

Second factor (F2), named as “Environmental Interactions Factor” explained 24.39 % of total variance and it was related to the variables of hardness, conductivity and Ca values of water and sediment. All parameters except Ca in sediment were strong positively loaded with this factor, and also parameter of Ca in sediment was moderate positively loaded with this factor.

Cluster analysis (CA) was used to determine the similarity groups between the stations. The diagram of CA calculated by using Mg, Ni, Mn and Cr levels of water, which were strongly correlated with each other at the 0.01 significant level, is given in Fig. 5. According to the CA, three statistically significant clusters were formed: Cluster 1 corresponded to S6 and S8 that were moderately contaminated by Cr and its compounds; Cluster 2 corresponded to S2, S3, S4, S1 and S7 that were uncontaminated by Cr and its compounds; Cluster 3 corresponded to S5 that was strongly contaminated by Cr and its compounds. The highest similarity was determined between S2 and S3 stations (99.53 %) and the lowest similarity was determined between S5 and S7 stations (38.33 %).

Accumulations of chromium and its compounds in water and sediment of S1, S2, S3 and S4 stations, which were located on the upside of Emet Stream and before falling into the Kinik Stream, showed that, these stations were not contaminated by these elements. But also it was determined that fishes of Emet Stream even caught from uncontaminated areas were adversely affected by chromium mines.

The S6 and S8 stations were located on the downside of Emet Stream and after the discharge of Kinik Stream. Sudden increase of Cr, Ni and Mn accumulations in water and sediment and also significant decrease of water and sediment quality were determined in these stations.

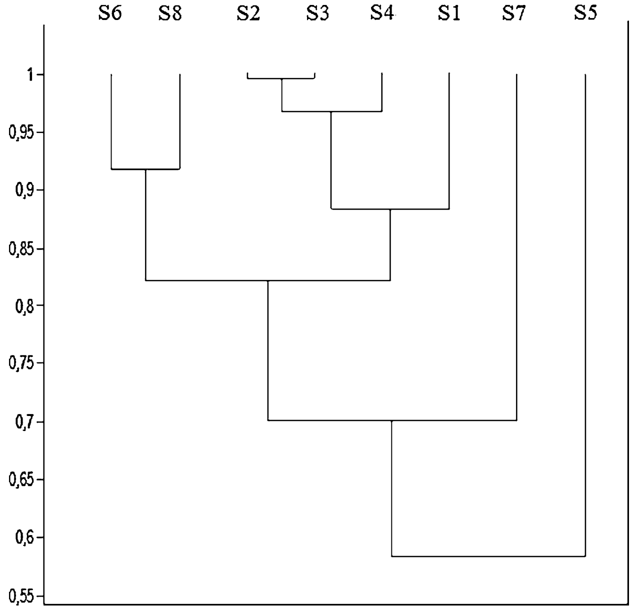

Fig. 5 Diagram of CA

Dursunbey Stream is an important branch of Emet Stream and S7 station was located on this stream. Although E7 station was located on the downside of Dursunbey Stream, it was not exposed to any chromium pollution. In addition, water and sediment quality of Emet Stream were increasing after the discharge of Dursunbey Stream in terms of Cr, Mn and Ni accumulations. So we can say that, Dursunbey Stream does not constitute any risk for Emet Stream Basin, on the contrary it is diluting the chromium, manganese and nickel levels in Emet Stream.

Turkey has a 6 % share of production in world's chromites mining and has a reserve of 25 million tons of chromium. Turkey's most important deposits of chrome are located in Harmancik, so this region has not only national but also global interest and importance (Uyanik 2007). Despite the global importance of the Harmancik region, it is the most important source of chromium pollution for the Emet Stream Basin and also Uluabat Lake. The wastes of Harmancik Chromium Mines are discharged into the Emet Stream through Kinik Stream and also into the Uluabat Lake through Emet Stream. The S5 station is located on the Kinik stream and is under pressure from the Harmancik chromium mines. The chromium levels of the S5 station were extremely high: an average of 153 times for water and 10 times for sediment than uncontaminated stations (S1, S2, S3, S4 and S7). So it can be understood that the transport of chromium in water is much faster than sedimentation because of the high flow rate and the alkaline water character of Kinik stream. Chromium is transported to Emet Stream and also Uluabat Lake directly before getting enough opportunity for sedimentation. According to data obtained, Kinik Stream is exposed to extreme chromium pollution and this adverse situation causes significant water and sediment quality problems for Emet Stream.

Conclusions

Emet Stream Basin is a mining area that has international importance. Therefore heavy metal pollution is a significant problem for the basin and Harancik Chromium Mines are the main inorganic pollution source for the ecosystem. The aim of this study was to evaluate the effects of chromium mines on abiotic and biotic components of the aquatic systems by using some mono and multi statistical techniques.

In general, extreme Cr and Ni accumulations both in water and sediment were identified in Kinik Stream, where the chromium mines located, and downsides of Emet Stream (after from the discharge of Kinik Stream). Significant positive correlations were determined between Cr accumulations with Mg, Ni and Mn in biotic and abiotic components of the basin. Transport of Cr and interactive elements in water were much faster than sedimentation because of high flow rate and alkaline water character of Kinik Stream. It was also determined that Dursunbey Stream, which is an important braches of Emet Stream, did not constitute any risk for the system in terms of Cr and Ni accumulations.

According to the results of FA, two factors which explain 83.53 % of the total variance named as "Chromium Mines Factor" and "Environmental Interactions Factor" were determined as affective factors on ecosystem quality. According to the results of CA, three statistically significant clusters were formed related with directly chromium mines.

It was revealed from the data that all fish species were adversely affected by the inorganic pollution and bioaccumulation levels of heavy metals in fishes of the basin were significantly higher that their environments. Cr and Ni bioaccumulations detected in liver and kidney tissue of *S. cii*, *C. tinca* and *B. oligolepis* were higher than detected in muscle and gill tissues. Cr and Ni levels in almost all fish tissues detected in S8 station (contaminated station) were significantly higher than detected in uncontaminated stations. Cr levels in muscle tissues of all fish species were much higher than the limit values for human consumption (higher an average of three times for *S. cii* and *B. oligolepis* and nine times for *C. tinca* than the limit value). This means that, inorganic pollution in the region is a significant threatening factor not only for ecosystem health but also for human health.

Briefly, Emet Stream Basin is dramatically under pressure of Harmancik Chromium Mines and these adverse effects pose an important risk factor for the aquatic life of the basin and public health of the region. Also it is thought to be that Cr and Ni elements in the Emet Stream Basin, which have high toxicity, could become a limiting factor for the population density of fishes in the future.

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